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(54) 19-Nor-vitamin D3 compounds with substituent at 2-position.

(57) The 2α and 2β-hydroxy as well as the 2α(3'-hydroxypropoxy)- and 2β(3'-hydroxypropoxy)- and 2α(benzyloxy)- analogs of 19-nor-1α,25-dihydroxyvitamin D_3 are disclosed. The two 2-hydroxy analogs showed in vivo calcium transport with little or no bone calcium mobilization ; the 2β-more than the 2α-analog. Both analogs induced differentiation of malignant cells. The two analogs thus show promise in the treatment of osteoporosis. The 2α-hydroxypropoxy analog showed a selective activity profile, combining high potency in inducing differentiation of malignant cells with very low or no bone calcification activity, a possible use in the treatment of malignancies.

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This invention relates to biologically active vitamin D_3 compounds. More specifically, the invention relates to 19-nor-analogs of 1α-hydroxylated vitamin D_3 compounds having a substituent at the 2-position in the A-ring.

Background and Summary of the Invention

The 1α-hydroxylated metabolites of vitamin D – most importantly 1α,25-dihydroxyvitamin D3 and 1α,25-dihydroxyvitamin D_2 -- are known as highly potent regulators of calcium homeostasis in animals and humans, and more recently their activity in cellular differentiation has also been established. V. Ostrem et al, Proc. Natl. Acad. Sci. USA, (1987), 84, 2610. As a consequence, many structural analogs of these metabolites, such as compounds with different side chain structures, different hydroxylation patterns, or different stereochemistry, have been prepared and tested. Important examples of such analogs are 1α-hydroxyvitamin D3, 1α-hydroxyvitamin D_2, various side chain fluorinated derivatives of 1α,25-dihydroxyvitamin D_3, and side chain homologated analogs. Several of these known compounds exhibit highly potent activity in vivo or in vitro, and some of these have been found to exhibit an interesting separation of activities in cell differentiation and calcium regulation. This difference in activity provides these compounds with advantageous therapeutic activity profiles and thus numerous of these compounds are in use, or have been proposed for use, in the treatment of a variety of diseases such as renal osteodystrophy, vitamin D-resistant rickets, osteoporosis, psoriasis, and certain malignancies.

Recently, a new class of vitamin D analogs has been discovered, i.e. the so-called 19-nor-vitamin D compounds. 19-Nor-vitamin D compounds are vitamin D analogs in which the ring A exocyclic methylene group (carbon 19) typical of all vitamin D compounds has been removed and replaced by two hydrogen atoms. Specifically, these compounds exhibit a selective activity profile with high potency in inducing cellular differentiation, and minimal bone calcification activity. Such a differential activity profile renders these compounds useful for the treatment of malignancies, or the treatment of various skin disorders. Two different methods of synthesis of these 19-nor-vitamin D analogs have been described (Perlman et al. Tetrahedron Letters 31, 1823 (1990); Perlman et al Tetrahedron Letters 32, 7663 (1991), and DeLuca et al U.S. Patent 5,086,191).

In U.S. Patent 4,666,634, 2β3-hydroxy and alkoxy analogs of 1α,25-dihydroxyvitamin D_3 have been described and examined as potential drugs for osteoporosis and as antitumor agents See also T. Okano et al, Biochem. and Biophys. Res. Comm., (1989) 163, 1444. However the new analogs also have many undesired side effects most notable of which is the potential development of hypercalcemia upon administration.

In a continuing effort to explore the new 19-nor class of pharmacologically important vitamin D analogs, the 2α- and 2β-hydroxy as well as the 2α(3'-hydroxypropoxy)- and the 2,β(3'-hydroxypropoxy)- and 2α(benzyloxy)-analogs of 19-nor-1α,25-dihydroxyvitamin D_3 have now been synthesized. The two 2-hydroxy analogs showed in vivo calcium transport with little or no bone calcium mobilization; the 2β- more than the 2a- analog. Both 2-hydroxy analogs induced differentiation of malignant cells. These two analogs thus show promise in the treatment of osteoporosis. The 2α-hydroxypropoxy analog showed a selective activity profile, combining high potency in inducing differentiation of malignant cells with very low bone calcium mobilizing activity, a possible use in the treatment of cancer.

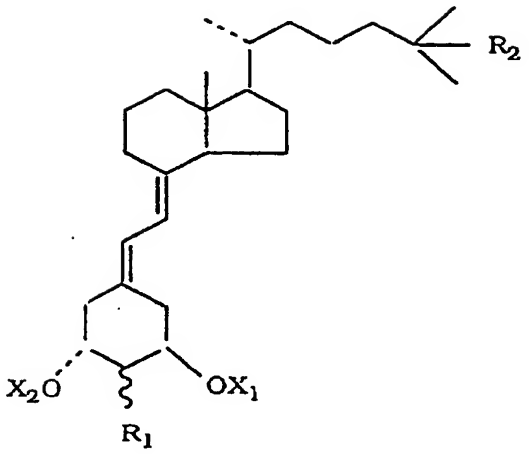
Brief Description of the Drawings

Figs. 1a and 1b are graphs of the percent differentiation of HL-60 cells versus concentration for a prior art vitamin D_3 compound and three of the new 19-nor-vitamin D_3 compounds; and

Figs. 2a and 2b are graphs of the competitive binding ability versus concentration for the same four compounds as in Figs. 1a and 1b.

Disclosure of the Invention

The new 19-nor-Vitamin D_3 analogs having a substituent at the 2-position in the A-ring are represented by the following general formula:



where X_1 and X_2 are each selected from hydrogen or a hydroxy protecting group, R_1 which may be in either α or β position is a hydroxy group, protected hydroxy group or the group OR_3 where R_3 is an alkyl, hydroxyalkyl, fluoroalkyl, arylalkyl or aryl group, and R_2 is hydrogen or a hydroxy group. Specific examples of the compounds represented by the formula above are: 1α,2α,25-trihydroxy-19-nor-vitamin D_3; 1α, 2β, 25-trihydroxy-19-nor-vitamin D_3; 1α,25-dihydroxy-2α-(3'-hydroxypropoxy)-19-nor-vitamin D_3; 1α,25-dihydroxy-2β-(3'-hydroxypropoxy)-19-nor-vitamin D_3 and 1α,25-dihydroxy-2α(benzyloxy)-19-nor-vitamin D_3.

The starting material for the synthesis of these new 19-nor compounds is the commercially available synthon (1R,3R,4R,5R) (-) quinic acid, D. Desmaele et al, Tetrahedron Letters (1985) 26, 4941, which after esterification with methanol in the presence of catalytic amount of p-toluene sulfonic acid, followed by treatment with tert-butyldimethylsilyl chloride and triethylamine in dimethyl formamide gave the protected methyl ester 1. Reduction with diisobutylaluminum hydride gave polyalcohol 2 followed by sodium periodate oxidation to the cyclohexanone derivative 3. The 4-hydroxy group was protected with N-(trimethylsilyl) imidazole to give 4.

The trimethylsilyl protecting group in this particular case with the very hindered 2-hydroxy group gave the needed chemoselectivity in the 2 position throughout all subsequent steps. Peterson reaction with methyl (trimethylsilyl)acetate in the presence of LDA in anhydrous tetrahydrofuran gave a mixture of the protected cyclohexylidene esters 5a, 5b. The latter was reduced to the allylic alcohols 6a, 6b with diisobutylaluminum hydride and separated and finally transformed to the desired phosphine oxide 7a and 7b by in situ conversion to the tosylate with BuLi and tosylchloride followed by lithium diphenylphosphide and oxidation with hydrogen peroxide. (During all these steps the 2 trimethylsilyloxy group was not removed).

The synthesis of the CD ring Windaus Grundmann ketone with the appropriate protected side chain is well documented in the literature. See E. G. Baggiolini et al, J. Org. Chem. (1986) 51, 3098; F. J. Sardina et al, J. Org. Chem. (1986) 51 1264; and U. S. Patent 4,804,502. In the present disclosure, the recently described procedure i.e. ozonolysis followed by RuO_4 oxidation of commercial vitamin D_3 was chosen. See J. Kiegiel et al, Tetrahedron Letters 32, 6057 (1991). The 25-OH of the Grundmann ketone was protected with TES-Cl, imidazole, in DMF to give chemoselectivity from the 2-OTMS group in the 19-nor product. With the required synthons on hand, the final convergent formation of 19-nor-1α,2α and -2β,25-dihydroxy-cholecalciferol 9a, 10a was then accomplished. See B. Lythgoe et al, J. Chem. Soc. Perkin Trans. 1, (1978) 590; B. Lythgoe et al, J. Chem. Soc. Perkin Trans 1. (1976) 2386; and H. T. Toh et al, J. Org. Chem. (1983), 48, 1414. Wittig Horner reaction of the lithium phosphinoxy carbanion prepared from 7a and n.butyl lithium at -78°C in tetrahydrofuran with the protected Windaus Grundmann ketone 8 proceeded to give the desired 19-nor-vitamin derivative 9a which after deprotection gave the crystalline 19-nor-1α,2α,25-trihydroxyvitamin D_3 10a; 10b was obtained in the same manner from 7b.

For the synthesis of the 19-nor-2α(3'-hydroxypropoxy)-1α,25-dihydroxyvitamin D_3 14a,9a was partially hydrolyzed under carefully controlled conditions: i.e. 9a was treated with a mixture of 8:8:1 tetrahydrofuran acetic acid water at room temperature for 4.5 h and the resulting mixture separated by HPLC to give the free 2α-hydroxy 11a. 3-bromo-1-tert.butyldimethylsilyloxy propane 12 was chosen as the alkylating agent and was prepared by silylation from the corresponding bromo alcohol. 11a was treated with sodium hydride and the protected bromo compound 12 in the presence of 18-Crown-6 in anh. DMF for 48 h to give the protected 13a which

was deprotected with Bu_4NF in THF to give the expected 14a; 14b was obtained in the same manner from 11b. Under the same conditions 11a with benzylbromide gave after deprotection 16a.

The stereochemistry at the 2-position of 5 was determined first by separation of the enantiomers followed by partial hydrolysis, and acylation. The two enantiomer's structure was then determined by NOE followed by 2D NMR in deuterated benzene. This revealed that under the reaction conditions the major component was the 2α-compound 5a and the minor the 2β-compound 5b (3:1).

As used in the description, and in the claims, the term "hydroxy-protecting group" refers to any group commonly used for the protection of hydroxy functions during subsequent reactions, including, for example, acyl or alkylsilyl groups such as trimethylsilyl, triethylsilyl, t-butyldimethylsilyl and analogous alkyl or arylsilyl radicals, or alkoxyalkyl groups such as methoxymethyl, ethoxymethyl, methoxyethoxymethyl, tetrahydrofuranyl or tetrahydropyranyl. A "protected-hydroxy" is a hydroxy function derivatized by one of the above hydroxy-protecting groupings. "Alkyl" represents a straight-chain or branched hydrocarbon radical of 1 to 10 carbons in all its isomeric forms, such as methyl, ethyl, propyl, isopropyl, butyl, isobutyl, pentyl, etc., and the terms "hydroxyalkyl," "fluoroalkyl" and "arylalkyl" refer to such an alkyl radical substituted by one or more hydroxy, fluoro or aryl groups respectively. An "acyl" group is an alkanoyl group of 1 to 6 carbons in all its isomeric forms, or an aroyl group, such as benzoyl, or halo-, nitro- or alkyl-substituted benzoyl groups, or an alkoxycarbonyl group of the type alkyl-O-CO-, such as methoxycarbonyl, ethoxycarbonyl, propyloxycarbonyl, etc., or a dicarboxylic acyl group such as oxalyl, malonyl, succinoyl, glutaroyl, or adipoyl. The term "aryl" signifies a phenyl-, or an alkyl-, nitro- or halo-substituted phenyl group. The term alkoxy signifies the group alkyl-O-. This invention is more specifically described by the following illustrative examples. In these examples specific products identified by Arabic numerals (e.g. 1, 2, 3, etc.) refer to the specific structures so identified in the preceding description and in the Schemes.

Example 1

Preparation of 1α,2α,25-trihydroxy-19-nor-vitamin D_3 (10a) and 1α,2β,25-trihydroxy-19-nor-vitamin D_3 (10b).

(a) (1R,3R,4R,5R)(-) Methyl Quinicate.

Referring first to Scheme I, p-toluene sulfonic acid (0.5g) was added to a solution of quinic acid (12.74 g, 66.3 mmol) in methanol. The solution was stirred for 24 h. Solid $NaHCO_3$ (1.0g) was added and after 15 min the solution was filtered and concentrated to give 12.61 g (61.16 mmol) of the methyl ester in 92% yield.

(b) (1R,3R,4R,5R) Methyl 3,5-Bis(tert-butyldimethylsilyloxy), 1,4-dihydroxycyclohexane-carboxylate (1).

tert-Butyldimethylsilyl chloride (6.73 g, 44.62 mmol) was added to a solution of methyl (1R,3R,4R,5R)(-) quinicate (3.68 g., 17.85 mmol) and triethyl amine (6.2 mL, 44.62 mmol) in 44 mL or anhydrous dimethyl formamide at 0°C with stirring. After 4 h the solution was warmed to room temperature and stirring continued for another 14 h. The solution was poured into water and extracted with ether. The combined organic layers were extracted with brine, dried over anhydrous $MgSO_4$, filtered and concentrated. The residue was purified by column chromatography on silica gel, eluting with 5-10% ethyl acetate in hexane mixtures, to give 4.6 g (60%) of 1 as a white solid. M.p. 82-82.5°C (after recrystallization from hexanes). ^1H NMR ($CDCl_3$, 500 MHz) δ 4.53 (bs, 1 H), 4.36 (bs, 1 H), 4.11 (ddd, 1 H), 3.76 (s, 3 H), 3.42 (dd, 1 H), 2.31 (bs, 1 H), 2.18 (bd, 1 H), 2.05 (ddd, 2 H), 1.82 (dd, 1 H), 0.91 (s, 9 H), 0.89 (s, 9H) 0.15 (s, 3 H), 0.14 (s, 3 H), 0.11 (s, 3 H), 0.09 (s, 3 H) MS m/e (relative intensity) 377 (70), 227 (91).

(c) (1R,3R,4R,5R) [3,5-Bis(tert,-butyldimethylsilyloxy)-1,4-dihydroxy]-1-hydroxymethylcyclohexane (2).

Diisobutyl aluminum hydride (45mL, 45.0 mmol, 1.0 M in hexanes) was added to a solution of the ester (3.26 g, 7.5 mmol) in ether (45 mL) at -78°C. After 20 min. the solution was warmed to -23°C and stirred for 2h. The solution was diluted with ether and then 2N potassium sodium tartrate was slowly added. The solution was warmed to room temperature and stirred for 15 min. The ether layer was separated and the aqueous layer extracted with ether. The combined ether layers were extracted with brine, dried over anh. $MgSO_4$, filtered and concentrated. The material was further purified by column chromatography on silica gel with 25% ethyl acetate/hexanes to give 83% of 2 (2.52 g, 6.20 mmol). Mp. 108-109°C from hexanes. ^1H NMR ($CDCl_3$, 500 MHz) δ 4.52 (bs, 1 H), 4.32 (bs, 1 H), 4.12 (ddd, 1 H), 3.40 (dd, 1 H), 3.33 (dd, 2 H), 2.28 (d, 1 H), 2.11 (dd, 1 H), 2.00 (ddd, 2 H), 1.52 (dd, 1 H), 1.33 (dd, 1 H), 0.91 (s, 9 H), 0.90 (s, 9 H), 0.16 (s, 3 H), 0.14 (s, 3 H), 0.12 (s,

3 H), 0.11 (s, 3 H), MS m/e (relative intensity) : 349 (8), 331 (13), 239 (12), 199 (100).

(d) (3R,5R) [3,5-Bis(tert.-butyldimethylsilyloxy) 4-hydroxy]-1-cyclohexanone (3).

Sodium periodate saturated water (28.5 mL) was added to the triol 2 (1.91 g, 4.7 mmol) in methanol (124 mL) at 0°C. The solution was stirred for 1 h, then poured into water and extracted with ether. The combined ether fractions were washed with brine, dried over anhydrous $MgSO_4$, filtered and concentrated, to give 1.72 g (4.59 mmol) of 3 (98%). No further purification was required. Mp. 98-100°C from hexanes. ^1H NMR ($CDCl_3$, 500 MHz) δ 4.28 (m. 2 H), 3.80 (bs, 1 H), 2.77 (dd, 1 H, J=14.3, 3.4 Hz), 2.59 (dd, 1 H, J=13.1, 10.7 Hz), 2.45 (dd, 1 H, J=14.1, 5.2 Hz), 2.25 (bd, 1 H, J=15.9 Hz), 0.90 (s, 9 H), 0.85 (s, 9 H), 0.08 (s, 3 H), 0.08 (s, 3 H), 0.06 (s, 6 H), MS m/e (relative intensity) 317 (62), 231 (16), 185 (76), 143 (100).

(e) (3R,5R) [3,5-Bis (tert.-butyldimethylsilyloxy)-4-tri-methylsilyloxy]-1-cyclohexanone (4).

N-(Trimethylsilyl)imidazole (2.52 mL, 26.67 mmol) was added to a solution of the ketoalcohol 3 (1.56 g, 4.167 mmol) in methylene chloride (38 mL). The solution was stirred for 20 h. Water (1 mL) was added and the solution stirred for 30 min. Brine and methylene chloride was added. The brine was extracted with methylene chloride. The combined methylene chloride fractions were dried with anh. $MgSO_4$, filtered and concentrated. The residue was further purified by column chromatography on silica gel with 10% ethyl acetate in hexane to give 4 (1.76 g, 3.95 mmol) in 95% yield. ^1H NMR ($CDCl_3$, 500 MHz) δ 4.25 (m, 1 H). 4.13 (m, 1 H), 4.04 (m, 1 H), 2.74 (ddd, 2 H), 2.38 (dd, 1 H), 2.19 (dd, 1 H), 0.90 (s, 9 H), 0.86 (s, 9 H), 0.16 (s, 9 H), 0.07 (bs, 12 H). MS m/e (relative intensity): 431 (5), 389 (100), 299 (45), 257 (28).

(f) (S)-and (R)-(3R,5R) Methyl [3,5-bis(tert.-butyldimethylsilyloxy-4-hydroxy]-cyclohexylidene carboxylate (5a and 5b).

n.Butyl lithium (2.3 mL, 3.0 mmol) 1.3 M in hexanes was added to a solution of diisopropylamine (0.42 mL, 3.0 mmol) in anhydrous tetrahydrofuran (2.0 mL) under argon at -78°C with stirring and methyl (trimethylsilyl) acetate (0.49 mL. 3.0 mmol) was added. After 15 min the protected keto compound 4 (0.629 g, 1.4 mmol) in anhydrous tetrahydrofuran (2.0 + 1 mL) was added. The solution was stirred for 2 h at -78°C. The reaction mixture was quenched with saturated ammonium chloride solution and extracted with ether. The combined ether fractions were washed with brine, water and dried over anhydrous $MgSO_4$, filtered and evaporated. The product was further purified by SepPak filtration in 5% ethyl acetate in hexane to give 0.693 g (98%) of a mixture of the two stereoisomer allylesters 5a and 5b. For analytical purposes the two allylesters were separated by HPLC (1% ethyl acetate in hexane, Zorbax Sil 10 x 25 cm, with differential refractometer as detector.)
5a Peak II (Major) (S): ^1H NMR ($CDCl_3$, 500 MHz) δ 0.04, 0.05, 0.08 (3H, 3H and 6H, each s, 4x SiMe), 0.13 (9H, s, $SiMe_3$), 0.86 (9H, s, Si-t-Bu), 0.89 (9H, s, Si-t-Bu), 2.00 (1H, dd, J=13.5, 4.7 Hz, 6β-H), 2.60 (1H, br d, J~13.5 Hz, 6α-H), 2.74 and 3.28 (1H and 1H, each br m, 2-H_2), 3.62 (1H, narrow m, 4β-H), 3.68 (3H, s, OMe), 3.86 (1H, ~q, J~4 Hz, 5α-H), 3.95 (1H, dt, J=9.5, 2.5 Hz, 3β-H), 5.63 (1H, s, 7-H).
5b Peak 1 (minor) (R): ^1H NMR ($CDCl_3$, 500 MHz) δ 0.04, 0.05, 0.06 (3H, 3H and 6H, each s, 4x SiMe), 0.13 (9H, s, $SiMe_3$), 0.84 (9H, s, Si-t-Bu), 0.89 (9H, s, Si-t-Bu), 2.12 (1H, dd, J=12.7, 3.8 Hz, 6α-H), 2.57 (1H, br t, J~12 Hz, 6β-H), 2.62 and 3.35 (1H and 1H, each br d, J~13 Hz, 2-H_2), 3.65 (1H, narrow m, 4α-H), 3.66 (3H, s, OMe), 3.86 (1H, ~q, J~4 Hz, 3β-H), 3.99 (1H, dt, J=9.9, 3.7 Hz, 5α-H), 5.70 (1H, s, 7-H).

(g) (S)- and (R)-(3R,5R) [3,5-Bis(tert.-butyldimethylsilyloxy) (4-trimethylsilyloxy)-cyclohexylidene] ethanol (6a and 6b).

A solution of 410 mg of mixture of esters 5a, 5b (0.82 mmol) in 8 mL of anhydrous toluene was treated at -78°C under argon with 7 mL (10.5 mmol) of a 1.5 M solution of diisobutylaluminum hydride in toluene. After the addition stirring was continued for 1 h at -78°C. The reaction mixture was then quenched by the addition of 2N potassium sodium tartrate, the organic phase was separated, and the aqueous phase extracted with ethyl acetate. The combined organic phases were washed with water and brine and dried over anhydrous $MgSO_4$, filtered and evaporated. The residue was purified by fast filtration through a silica gel column, using 20% ethyl acetate in hexane as eluent, to give 352 mg (91%) alcohols 6a and 6b which were separated by HPLC, Zorbax Sil 10 x 25 cm column, 10% ethyl acetate in hexane. First eluted the minor 4R alcohol (42 mg) 6b followed by the major 4S alcohol 6a (188 mg) (61% recovery).
6a Peak II (major) (S): ^1H NMR ($CDCl_3$, 500 MHz) d 0.04, 0.05, 0.06, 0.07 (3H, each s, 4x SiMe), 0.13 (9H, s, $SiMe_3$), 0.87 and 0.89 (9H and 9H, each s, 2x Si-t-Bu), 1.93 (1H, dd, J=13.5, 5.5 Hz, 6β-H), 2.24 (1H, br d,

J~12.6 Hz, 2β-H), 2.38 (1H, dd, J=12.6 Hz, 9.2 Hz, 2α-H), 2.50 (1H, dd, J=13.5, 3.5 Hz, 6α-H) 3.57 (1H, narrow m, 4β-H), 3.80 (1H, dt, J=3.5 Hz, 5.5 Hz, 5α-H) 3.89 (1H, ddd, J=9.2, 3.0, 2.6 Hz, 3β-H, 4.13 (2H, m, 8-H_2), 5.45 (1H, t, J=6.9 Hz, 7-H).

6b Peak I (minor) (R): ^{1}H NMR ($CDCl_3$, 500 MHz) δ 0.06, 0.07 (6H and 6H, each s, 2x $SiMe_2$), 0.13 (9H, s, $SiMe_3$), 0.87 and 0.89 (9H and 9H, each s. 2x Si-t-Bu), 2.06(1H, dd, J=12.3, 3.9 Hz, 6α-H), 2.23 (1H, dd, J=13.6, 4.0 Hz, one of 2-H_2), 2.38 (1H, br d, J=13.6 Hz, one of 2-H_2), 2.45 (1H, br t, J~11 Hz, 6β-H), 3.62 (1H, dd, 4.0 and 2.0 Hz, 4α-H), 3.82 (1H, ~q, J~4 Hz, 3β-H), 3.89 (1H, ddd, J=10, 4 and 2 Hz, 5α-H), 4.03 and 4.12 (1H and 1H, each br m, 8-H_2), 5.57 (1H, t, J=7.1 Hz, 7-H).

(h) (S)- and (R)-(3R,5R)-[3,5-Bis(tert.-butyldimethylsilyloxy-4-trimethylsilyloxy)-cyclohexylidene] ethyl diphenylphosphine oxide (7a and 7b).

265 mg dry allylalcohol 6a (0.56 mmol) was dissolved in 5.0 mL anhydrous tetrahydrofuran and 0.35 mL (0.56 mmol) n. Butyl lithium (1.6 M in hexanes) added under argon. 106 mg (0.56 mmol) recrystallized dry tosylchloride was dissolved in 1.0 mL anhydrous tetrahydrofuran and added to the allylalcohol-BuLi solution under argon at 0°C. The solution was stirred at 0°C for 5 min. and set aside at 0°C.
In another dry flask with air replaced by argon 350 µl (0.56 mmol) n.Butyl lithium (1.6 M in hexanes) was added to 100 µl (0.56 mmol) diphenylphosphine in 200 µl anhydrous tetrahydrofuran at 0°C with stirring under argon. To the orange solution was syphoned under argon pressure at 0°C the tetrahydrofuran solution of the allylic tosylate. The resulting orange solution was stirred an additional 30 min at 0°C and quenched by the addition of water. Solvents were evaporated under reduced pressure and the residue was dissolved in 5 mL of dichloromethane. The dichloromethane layer was stirred with 20 mL of 10% hydrogen peroxide at 0°C for 1 h. The dichloromethane layer was separated and washed with cold aqueous sodium sulfite, water and brine, dried over anhydrous $MgSO_4$, filtered and evaporated. The residue was dissolved in 20% 2-propanol in hexane and passed through a silica SepPak and purified by HPLC (Zorbax-Sil 9.4 x 25 cm column, 20% 2-propanol in hexane) to give 220 mg (60%) of the crystalline phosphine oxide 7a.
7a UV(EtOH): λ_{max} 258, 265, 272 nm. ^{1}H NMR ($CDCl_3$, 500 MHz) δ - 0.02, 0.00, 0.01, 0.03 (3H, each s, 4 x SiMe), 0.09 (9H, s, $SiMe_3$), 0.83 and 0.87 (9H and 9H, each s, 2x Si-t-Bu). 1.86 (1H, br d, J~13.5 Hz, one of 6-H_2), 1.99 and 2.08 (1H and 1H, each m, 2-H_2), 2.42 (1H, br d, J~13.5 Hz, one of 6-H_2), 3.10 (2H, m, 8-H_2), 3.51 (1H, narrow m, 4β-H), 3.72 (1H, dt, J=3.7, 5.2 Hz, 5αH), 3.81 (1H, ddd, J=8.8, 4.2, 2.4 Hz, 3β-H), 5.24 (1H, q, J=6.9 Hz, 7-H), 7.46, 7.52 and 7.71 (4H, 2H and 4H, each m, Ar-H). Mass spectrum (exact mass calcd for $C_{36}H_{58}O_4Si_3P$ 658.3459 found 658.3453), m/e (relative intensity) 658 (M^+, 1), 643 (3), 601 (100), 526 (12), 469 (43).
7b was prepared in the same way as 7a except from the corresponding 6b.
7b ^{1}H NMR ($CDCl_3$, 500 MHz) δ 0.02, (12 H, s, 2x SiMe), 0.09 (9H, s, $SiMe_3$), 0.85 (18 H, s, 2x Si-t-Bu), 1.88 (1H, br d, J~14 Hz, one of 2-H_2), 2.01 (1H, br d J~12 Hz, 6α-H), 2.06 (1H, br d, J~14 Hz, one of 2-H_2), 2.34 (1H, br m, 6β-H) 3.02 and 3.14 (1 H and 1 H, each br m, 8-H_2), 3.51 (1H, narrow m, 4α-H), 3.71 (1H, ~q, J=4.4 Hz, 3β-H), 3.84 (1H, m, 5α-H), 5.27 (1H, m, 7-H), 7.46, 7.52 and 7.71 (4H, 2H and 4H, each m, Ar-H).

(i) Triethylsilyloxy Grundmann Ketone (8).

Referring now to Scheme II, the CD-ring fragment for the 19-nor vitamin D-derivative was prepared by ozonolysis of commercial vitamin D_3 to provide 20 followed by RuO_4 oxidation to give the 25-hydroxy-Grundmann ketone 21. To 30 mg ketone 21 (0.1 mmol) and 28 mg Imidazole (0.41 mmol) in 500 µl anhydrous dimethyl formamide was added 40 µl triethylsilyl chloride (0.24 mmol). The mixture was stirred at room temperature for 2 h. Ethyl acetate was added and water, and separated. The ethyl acetate layer was washed with water, brine, dried over anh. $MgSO_4$, filtered and evaporated. The residue was passed through a silica gel SepPak column in 10% ethyl acetate in hexane, and after evaporation purified by HPLC (Zorbax SII 9.4 x 25 cm column, 10% ethyl acetate in hexane, using a differential refractometer) to give 31 mg (79%) of the pure protected ketone 8.
^{1}H NMR ($CDCl_3$, 500 MHz) δ 0.56 (6H, q, J=8.0 Hz, 3 x Si-CH_2), 0.64 (3H, s, 18-H_3), 0.94 (9H, t, J=8.0 Hz, 3 x SiEt), 0.95 (3H, d, J=6.5 Hz, 21-H_3), 1.19 (6H, br s, 26- and 27-H_3), 2.45 (1H, dd, J=11.7, 7.4 Hz, 14α-H).

(j) 1α,2α,25-trihydroxy-19-nor-vitamin D_3 (10a).

16.9 mg (25.7 µmol) phosphine oxide 7a was dissolved in 200 µl anhydrous tetrahydrofuran, cooled to 0°C and 20 µl (26 µmol) n.butyl lithium (1.3 molar in hexanes) added under argon with stirring. The solution turns deep orange. The mixture was cooled to -78°C and 7.5 mg (21 µmol) protected ketone 8 added in 200

µl + 100 µl anhydrous tetrahydrofuran. The mixture was stirred under argon at -78°C for 1 h (at that time the solution became colorless) and at RT 18 hr. Ethyl acetate was added and the organic phase washed with water, brine, dried over anhydrous $MgSO_4$, filtered and evaporated. The residue was dissolved in 10% ethyl acetate in hexane, passed through a silica SepPak and washed with 40 mL of the same to give the 19-nor vitamin D derivative 9a. The Sep Pak was then washed with 20% 2-propanol in hexane to recover 5 mg unchanged diphenylphosphine oxide. 9a was purified by HPLC in 10% ethyl acetate in hexane (Zorbax Sil 9.4 x 25 cm column) to give 8.2 mg of the protected 19-nor vitamin D_3 derivative 9a (54%).

^1H NMR ($CDCl_3$, 500 MHz) δ 0.04, 0.05, 0.06 (3H, 3H, and 6H, each s, 4x SiMe), 0.12 (9H, s, $SiMe_3$), 0.55 (3H, s, 18-H_3), 0.56 (6H, q J=7.4 Hz, 3 x $SiCH_2$), 0.87 and 0.88 (9H and 9H, each s, 2 x Si-t-Bu), 0.92 (3H, d, J=6.1 Hz, 21-H_3), 0.95 (9H, t, J=7.4 Hz, 3 x SiEt), 1.19 (6H, br s, 26-and 27-CH_3), 2.79 (1H, br d, J=12.6 Hz, 9β-H), 3.53 (1H, m, 2β-H), 3.80 (1 H, m, 3α-H), 3.88 (1 H, m, 1β-H), 5.81 and 6.10 (1 H and 1 H, each d, J=11.4 Hz, 6- and 7-H). Mass spectrum (exact mass calcd for $C_{47}H_{94}O_4Si_4$ 834.6229, found 834.6241) m/e (relative intensity) 834 (12), 805 (3), 702 (100), 645 (18), 599 (45).

All of 9a was dissolved in 1.0 ml of anhydrous tetrahydrofuran and treated with 150 µl of a 1M solution of tetrabutylammonium fluoride in tetrahydrofuran. The mixture was stirred under argon at RT for 16 h and extracted with ethyl acetate. The organic phase was washed with 10% $NaHCO_3$ solution, brine and dried over anhydrous $MgSO_4$, filtered and evaporated. The residue was filtered through a Silica SepPak and purified by HPLC (Zorbax Sil 9.4 x 25 cm column, 30% 2-propanol in hexane) to give 1.48 mg of pure 1α,2α,25-trihydroxy-19-nor-vitamin D_3 10a.

UV (in EtOH) λ_{max}: 243, 251.5, 261 nm. Mass spectrum m/e (relative intensity): 420 (M^+, 100), 402 (55), 384 (18), 245 (45), 95 (95). 59 (95).

10a (2α-OH): ^1H NMR ($CDCl_3$, 500 MHz) δ 0.55 (3H, s, 18-H_3), 0.94 (3H, d, J=6.5 Hz, 21-H_3), 1.22 (6H, s, 26- and 27-CH_3), 2.62 (1H, dd, J=13.0, 4.2 Hz), 2.79 (1H, br d, J=12.9 Hz), 2.90 (1H, dd, J=14.3, 4.2 Hz), 3.25 (1H, br m), 3.53 (1H, dd, J=8.4, 2.6 Hz), 3.79 (1 H, br m), 4.10 (1 H, m), 5.81 and 6.38 (1 H and 1 H, each d, J=11.0 Hz. 6-and 7-H).

10b was prepared in the same way as 10a except from the corresponding 7b (Structure not shown).

10b (2β-OH): ^1H NMR ($CDCl_3$, 500 MHz) δ 0.55 (3H,s, 18-H_3), 0.94 (3H, d, J=6.8 Hz, 21-H_3), 1.22 (6H, s, 26- and 27-CH_3), 2.79 (1H, br d, J=13.0 Hz), 3.36 (2H, br m), 3.49 (1H, m), 3.68 (1 H, br m), 4.08 (1 H, m), 5.84 and 6.29 (1 H and 1 H, each d. J=11.3 Hz, 6- and 7-H).

Example 2

Scheme III illustrates the preparation of 1α,25-dihydroxy-2α(3'-hydroxypropoxy)-19-nor-vitamin D_3 (14a) and 1α,25-dihydroxy-2α(benzyloxy)-19-nor-vitamin D_3 (16a).

(a) 19-Nor-(1α,3β)-[bis(tert.-butyldimethylsilyloxy)-25-triethylsilyloxy-2α-hydroxyvitamin D_3 (11a).

10 mg 9a was stirred for 4.5 h at room temperature with 3 ml of a mixture of tetrahydrofuran-acetic acid-water (8:8:1). Ethyl acetate was added and washed with ice cold water, ice cold 10% $NaHCO_3$ solution until neutral, water and brine, and dried over anh. $MgSO_4$, filtered and evaporated. The residue was purified by HPLC (Zorbax Sil 10 x 25 cm column, 5% ethyl acetate in hexane) to give (in order of peak elutions) 1.2 mg unchanged starting material (12%), 3.5 mg of the expected 2-hydroxy compound 11a (38%) and 0.75 mg of 25-hydroxy compound (10%) (total recovery 60%).

11a (2α-OH): ^1H NMR ($CDCl_3$, 500 MHz) δ 0.06, 0.07, 0.08, 0.10 (each 3H, each s, 4x SiMe), 0.55 (3H, s, 18-H_3), 0.56 (6H,q, J=7.8 Hz, 3x $SiCH_2$) 0.87 and 0.88 (9H and 9H, each s, 2 x Si-t-Bu), 0.93 (3H, d, J=6.4 Hz, 21-H_3), 0.95 (9H, t, J=7.8 Hz, 3 x SiEt), 1.19 (6H, br s, 26- and 27-CH_3), 2.27 (1H, d, J=3.3 Hz, OH), 2.79 (1H, dd, J=12.3, 4.2 Hz, 9β-H), 3.51 (1H,dt, J~6 and 3 Hz; after D_2O : dd, J=5.8, 2.7 Hz, 2β-H), 3.91 (1H, dt, J=4.3, 5.8 Hz, 3α-H), 4.00 (1H,~dt, J~7.5, 3 Hz, 1β-H), 5.80 and 6.16 (1H and 1H, each, d, J=11.2 Hz, 6- and 7-H).

19-Nor-(1α,3β)-[bis (tert.-butyldimethylsilyloxy)-25-triethylsilyloxy-2β-hydroxy-vitamin D_3 (11b)

11b (not shown) was prepared the same was as 11a except from 9b (not shown)

11b (2β-OH): ^1H NMR ($CDCl_3$, 500 MHz) δ 0.06, 0.07, 0.08, 0.10 (each 3H, each s, 4x SiMe), 0.54 (3H, s, 18-H_3), 0.56 (6H, q, J=8.0 Hz, 3x $SiCH_2$), 0.86 and 0.89 (9H and 9H, each s, 2x Si-t-Bu), 0.93 (3H, d, J=6 Hz, 21-H_3), 0.94 (9H, t, J=8.0 Hz, SiEt), 1.19 (6H, br s, 26- and 27-CH_3), 2.81 (1H, br d, J~13 Hz, 9β-H), 3.59 (1H, narrow m, after D_2O : dd, J=3.6, 3.3 Hz, 2α-H), 4.00 (2H, m, 1β- and 3α-H), 5.80 and 6.19 (1H and 1H, each d, J=11.0 Hz, 6- and 7-H).

(b) 3-Bromo-1-(tert.-butyldimethylsilyloxy)-propane (12).

1.4 g (1 mmol) 3-bromo-1-propanol was dissolved in 5 ml of anhydrous dimethyl formamide and 3.0 g of imidazole, followed by 3.3 g of tert.-butyldimethylsilyl chloride was added at 0°C with stirring. The mixture was stirred at room temperature for 2 h. Ether was added, and the ether phase washed with water and brine. dried over anh. $MgSO_4$, filtered and evaporated. The residue was passed through a small silica gel column and eluted with hexane to give 2.03 g (80%) pure 12.
^1H NMR ($CDCl_3$, 500 MHz) δ 0.06 (6 H,s, $SiMe_2$), 0.90 (9H, s, Si-t-Bu), 2.03 (2H, ~ quint., J~6 Hz, C-CH_2-C), 3.51 (2H, t, J=6.2 Hz, CH_2-0), 3.73 (2H, t, J=5.8 Hz, CH_2-Br).

(c) 19-Nor-2α(3'-hydroxypropoxy)-1-dihydroxyvitamin D_3 (14a).

1.6 mg (2 µmol) 11a was dissolved in 200 µl of anh. dimethyl formamide and 3 mg sodium hydride (as 60% oil dispersion) followed by 3 mg 18-Crown-6 and 5 µl of the bromo compound 12 was added and the mixture stirred under argon atmosphere for 48 h. The mixture was extracted with ethyl acetate, washed with water, dried over anh. $MgSO_4$, filtered and evaporated. The residue was passed through a silica gel SepPak column and evaporated in AcOEt.
13a: ^1H NMR ($CDCl_3$, 500 MHz) δ 0.04, 0.05, 0.06, 0.07 (6H, 6H, 3H, 3H, each s, 6x SiMe), 0.55 (3H, s, 18-H_3), 0.56 (6H, q, J=7.5 Hz, 3x $SiCH_2$) 0.87 and 0.88 and 0.89 (9H, 9H and 9H, each s, 3x Si-t-Bu), 0.93 (3H, d, J=6 Hz, 21-H_3), 0.95 (9H, t, J=7.5 Hz, 3 x SIEt), 1.19 (6H, br s, 26-and 27-CH_3), 2.79 (1H, br d, J=14 Hz), 3.12 (1H, m), 3.4-4.1 (at least 7H, complex m), 5.80 and 6.12 (1H and 1H, each d, J=11 Hz, 6- and 7-H).
The residue was dissolved in 1 ml of anh. tetrahydrofuran and 0.5 ml tetrabutylammonium fluoride (1.0 M in THF) was added and stirred under argon atmosphere for 20 h. The residue was extracted with ethyl acetate, washed with water and 10% $NaHCO_3$ solution, water and brine, and dried over anh. $MgSO_4$, filtered and evaporated. The residue was passed through a silica gel SepPak column in 1:1 2-PrOH-hexane and purified by HPLC (Zorbax Sil 10mm x 25 cm column, 40% 2-PrOH-hexane) to give 202 µg of the expected product 14a (overall yield from 11a 21%) and 20 µg of 10a.
14a UV (in EtOH) λ_{max}: 243, 251.5, 261 nm. Mass spectrum (exact mass calcd for $C_{29}H_{50}O_5$ 478.3658, found 478.3659), m/e (relative intensity) 478 (M+, 5), 460 (6), 442 (2), 402 (4), 384 (3), 245 (15), 184 (20), 142 (100), 95 (50), 59 (38). ^1H NMR ($CDCl_3$, 500 MHz) δ 0.55 (3H, s, 18-H_3), 0.93 (3H, d, J=6.8 Hz, 21-H_3), 1.22 (6H, s, 26-and 27-H_3), 3.3-4.2 (at least 7H, complex m), 5.83 and 6.34 (1H and 1H, each d, J=11.2 Hz, 6- and 7-H).

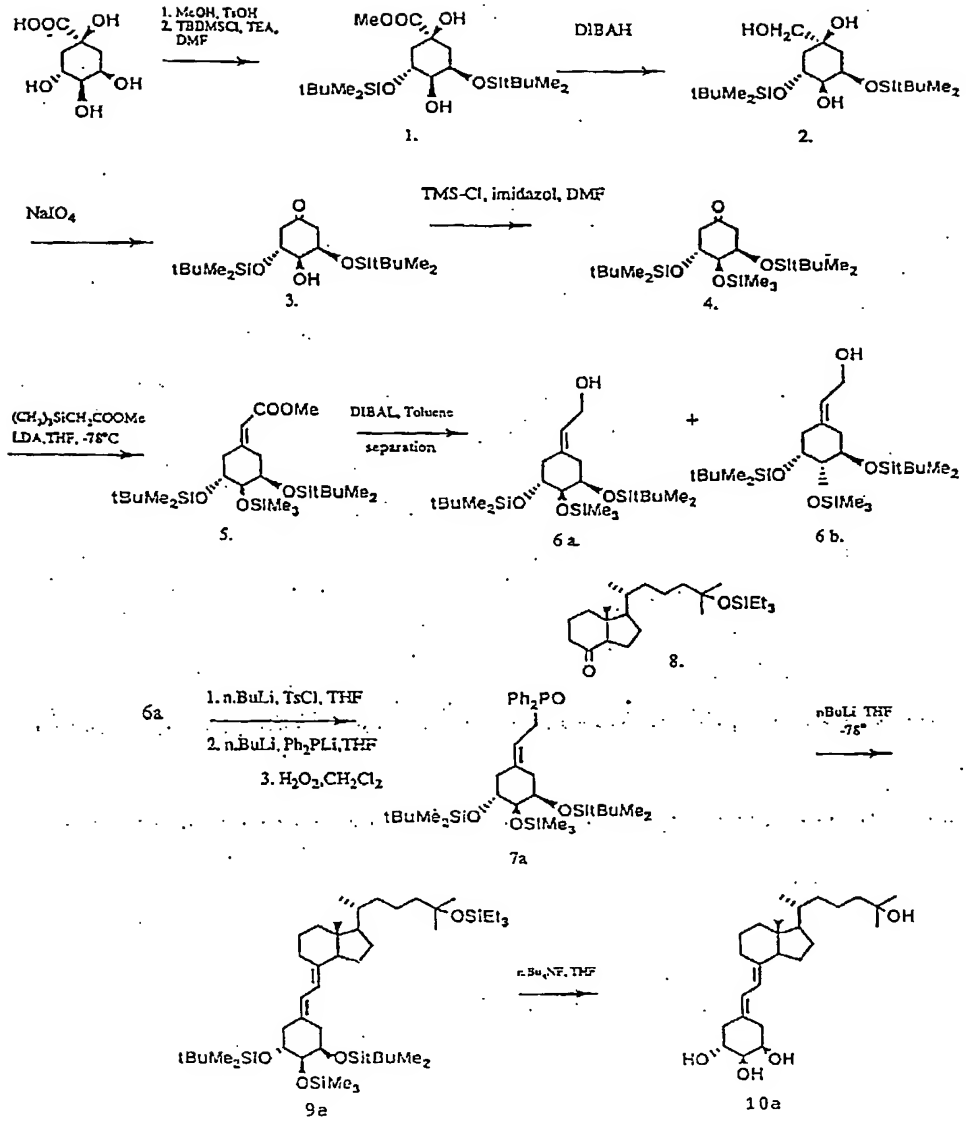
(d) 19-Nor-2β(3'-hydroxypropoxy)-1α,25-dihydroxyvitamin D_3 14b) (Structure not shown)

14b was prepared in the same way as 14a except from the corresponding 11b (structure not shown).
14b UV (in EtOH) λ_{max}: 243, 251.5, 261 nm. Mass spectrum m/e (relative intensity) 478 (M+, 5), 460 (20), 442 (18), 424 (4), 384 (3), 245 (28), 181 (28), 95 (50), 69 (100), 59 (68). ^1H NMR ($CDCl_3$, 500 MHz) δ 0.54 (3H, s, 18-H_3), 0.94 (3H, d, J=6.5 Hz, 21-H_3), 1.22 (6H, s, 26-and 27-H_3), 2.79 (1H, br d, J~12Hz, 9β-H), 3.0-4.3 (at least 7H, complex m), 5.83 and 6.29 (1H and 1H, each d, J=11.2 Hz, 6- and 7-H).

(e) 19-Nor-2α(benzyloxy)-1α,25-dihydroxyvitamin D_3 (16a).

1.6 mg (2 µmol) (11a) was dissolved in 200 µl of anh. dimethyl formamide and 3 mg sodium hydride (as 60% oil dispersion) followed by 3 mg 18-Crown-6 and 6 µl of benzylbromide benzene solution, (prepared from 120 µl benzylbromide in 1 mL benzene) was added and the mixture stirred under argon atmosphere for 48 h. The mixture was extracted with ethyl acetate, washed with water, dried over anh. $MgSO_4$, filtered and evaporated. The residue was passed through a silica gel SepPak column, evaporated to give 860 µg crude 15a, which was without purification deprotected. 860 µg crude 15a was dissolved in 200 µL of methanol and 10 mg methanol washed AG-50W-X4 cation exchange resin added. The mixture was stirred under argon atmosphere at RT for 18 h, filtered through a SepPak Silica cartridge and washed with 2-propanol. The solvent was evaporated under reduced pressure and the residue purified by HPLC (Zorbax Sil 10 mm x 25 cm column, 30% 2-PrOH-hexane mixture) to give 170 µg of the 2α-benzyloxy-compound mixture) to give 170 µg of the 2α-benzyloxy-compound 16a.
16a : UV (in EtOH) λ_{max}: 243, 251.5, 261 nm. Mass spectrum (exact mass calcd for $C_{33}H_{50}O_4$ 510.3709 found 510.3703), m/e relative intensity 510 (11), 492 (8), 474 (2), 401 (8), 91 (100). ^1H NMR ($CDCl_3$, 500 MHz) δ 0.55 (3 H, s, 18-H_3), 0.93 (3 H, d, J=6.7 Hz, 21-H_3), 1.22 (6H, s, 26- and 27-H_3), 2.79 (2H, m), 3.45 (1 H, dd, J=7.3, 3.0 Hz, 2β-H), 3.97 (1 H, m, 3α-H), 4.11 (1 H, m, 1β-H), 4.65 and 4.72 (1 H and 1 H, each d, J=11.8 Hz, O-CH_2'), 5.83 and 6.33 (1 H and 1 H, each d, J=11.2 Hz, 6- and 7-H), 7.2-7.4 (5H, br m, Ar-H).

SCHEME I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TMS-Cl, imidazol, DMF
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LDA, THF, -78°C
DIBAL, Toluene
separation
1.
2.
3.
4.
5.
6a.
6b.
8.
6a
1. n.BuLi, TsCl, THF
2. n.BuLi, Ph2PLi, THF
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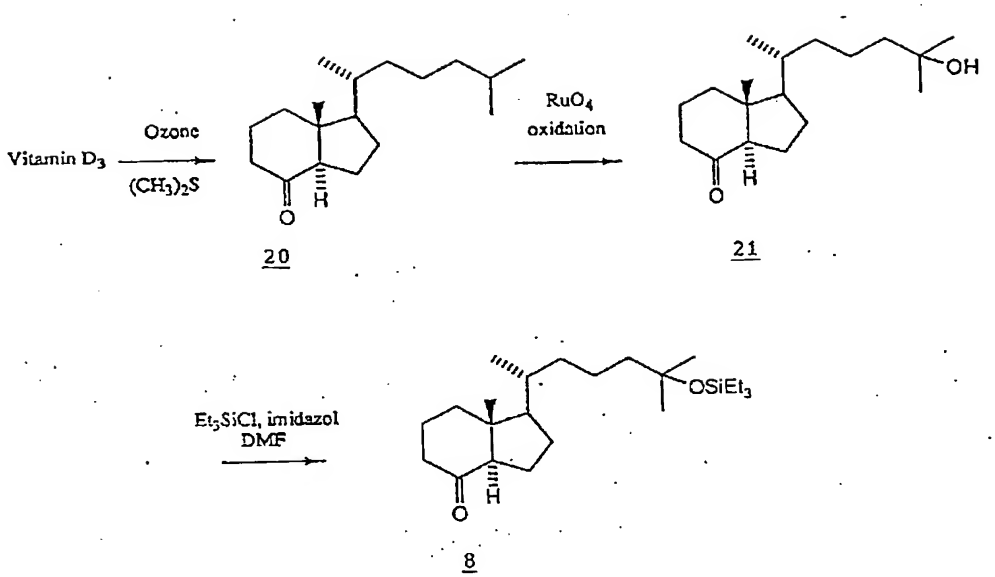
7a
n.Bu4NF, THF
9a
10a

SCHEME II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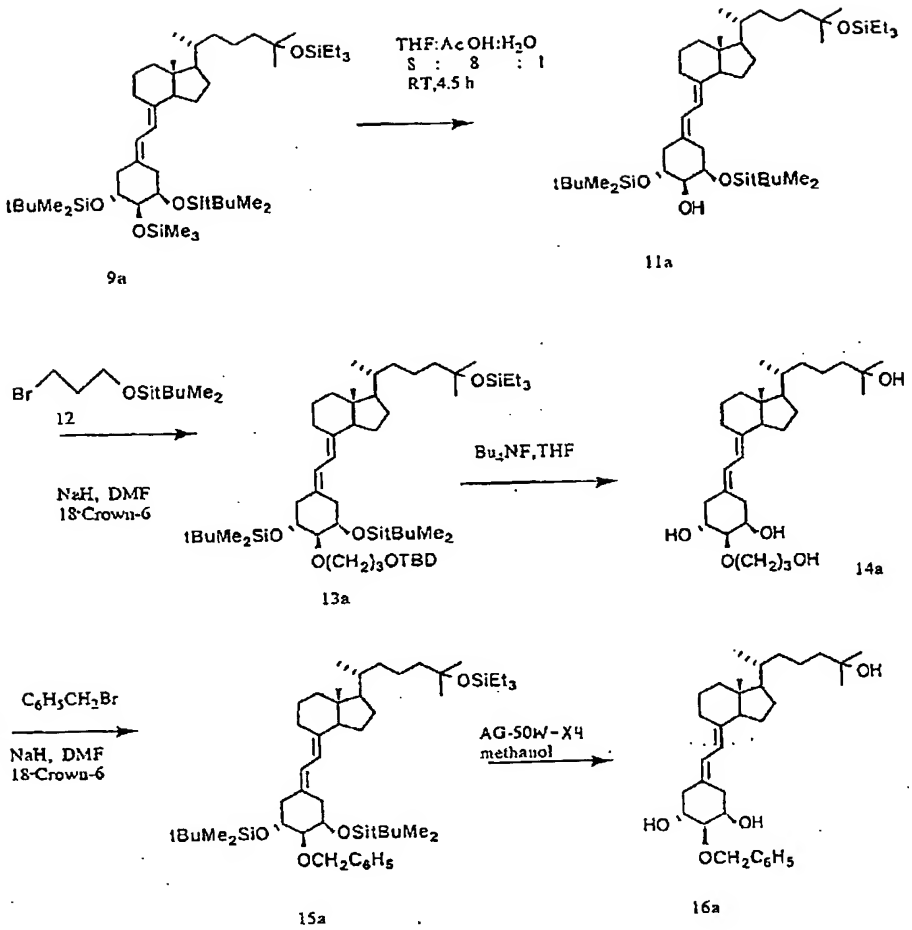
8

SCHEME III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Br
OSitBuMe2
12
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Bu4NF,THF
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13a
OH
HO
O(CH2)3OH
14a
11a
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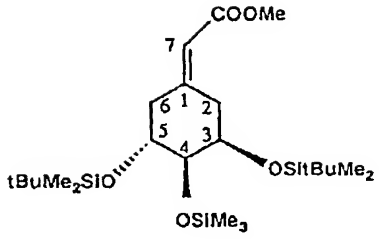
16a

SCHEME IV



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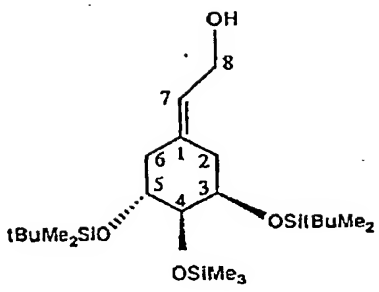

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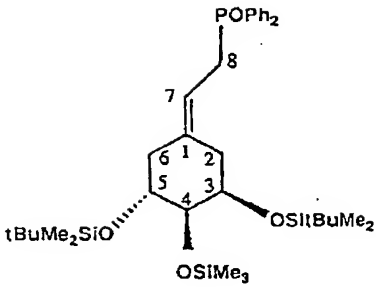
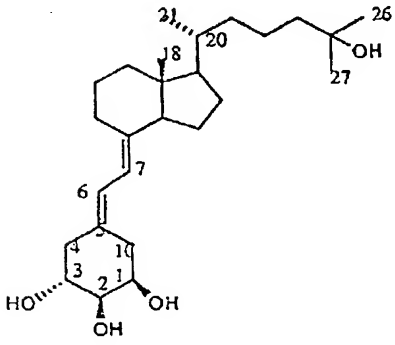


7a



10a

Biological Activity of 1α-Hydroxy-19-Nor Vitamin D Compounds

The novel compounds of this invention exhibit an unexpected pattern of biological activity. All of the 19-nor compounds exhibited high potency in promoting the differentiation of malignant cells. The two 2-hydroxy analogs showed in vivo calcium transport with little or no bone calcium mobilization; the 2β- more than the 2α- analog; while the 2α-hydroxypropoxy analog showed a selective activity profile, combining high potency in inducing differentiation of malignant cells with very low bone calcium mobilizing activity. This is illustrated by the biological assay results obtained for the claimed 19-nor-vitamin D_3 compounds, which are summarized in Figs. 1 and 2, and Table 1. Fig. 1 shows a comparison of the activity of the known active metabolite 1α,25-dihydroxyvitamin D_3 and two of the 19-nor analogs in inducing the differentiation of human leukemia cells (HL-60 cells) in culture to normal cells (monocytes). Differentiation activity was assessed by a standard differentiation assay, abbreviated in Fig. 1 as NBT (nitroblue tetrazolium reduction). The assay was conducted according to known procedures, as given, for example, by DeLuca et al U. S. Patent 4,717,721 and Ostrem et al, J. Biol. Chem. 262, 14164, 1987. For the assay, the differentiation activity of the test compounds is expressed in terms of the percent of HL-60 cells having differentiated to normal cells in response to a given concentration of test compound.

The results summarized in Fig. 1 clearly show that the new analogs, 1α,2α,25-trihydroxy-19-nor-vitamin D_3 and 1α,25-dihydroxy-2α-(3'-hydroxypropoxy)-19-nor-vitamin D_3, are as potent as 1α,25-dihydroxyvitamin D_3 in promoting the differentiation of leukemia cells. Thus in NBT assay close to 90% of the cells are induced to differentiation by 1α,25-dihydroxyvitamin D_3 at a concentration of 1×10^{-7} molar, and the same degree of differentiation is achieved by the two 19-nor analogs.

Fig. 2 shows a comparison of the same three compounds as in Fig. 1 illustrating their relative activity with regard to competitive binding to the vitamin D receptor. The competitive receptor binding was done with pig nuclear extract as described in Perlman et al. Biochemistry 29, 190-196 (1990) using the porcine extract prepared as described by Dame et al PNAS 82, 7825-7829 (1985). These data are used to demonstrate that the compounds described herein have relatively high in vivo activity, and have somewhat less activity than 1,25-$(OH)_2D_3$ in binding to the vitamin D receptor.

In regard to the biological data on the calcemic activity of these compounds reported in Table 1, Holtzmann weanling rats were maintained on 0.47% Ca, 3% P, for one week, then switched to a low Ca diet (0.02% Ca) for an additional three weeks. During the 4th week all animals were dosed with the appropriate compounds via the peritonal cavity. All doses were suspended in ethanol propylene glycol (5/95) and administered daily for seven days. None of the compounds produced hypercalcemia over the seven day dosing period. The 19-nor-2α-hydroxypropoxy-1α,25-dihydroxyvitamin D_3 did mobilize small amounts of Ca from bone at 130 or 325 pmole daily. However, these levels could easily be controlled by lowering the dose and the activity in this regard was far below that of the standard compound, 1,25 $(OH)_2D_3$.

The data in Table 1 illustrates that 19-nor-1α,2α,25-trihydroxyvitamin D_3 has biological activity in intestinal transport similar to that of 1,25-$(OH)_2D_3$ but possesses little or no bone calcium mobilizing activity even when given at 325 pmol/day. Similarly, the 1α,2β,25-trihydroxyvitamin D_3 compound has considerable activity in intestinal calcium transport but again lacks bone calcium mobilizing activity. On the other hand, the 19-nor-2α-hydroxypropoxy-1α,25-dihydroxyvitamin D_3 compound has an activity profile similar to 1,25-$(OH)_2D_3$ but has preferential activity on bone calcium mobilization.

The 2-propoxy compound would be useful in circumstances where an increased bone turnover is desirable such as low bone turnover osteoporosis. The other two compounds would find utility as a treatment for post-menopausal and senile osteoporosis because of their low bone calcium mobilizing activity with normal differentiative activity, normal binding to the receptor, and normal calcium transport activity. The compounds that could be considered for anti-cancer or psoriasis might be the 2α- and 2β-hydroxy compounds because of their tendency to not cause hypercalcemia.

For treatment purposes, the novel compounds of this invention can be formulated as solutions in innocuous solvents, or as emulsions, suspensions or dispersions in suitable innocuous solvents or carriers, or as pills, tablets or capsules, containing solid carriers according to conventional methods known in the art. For topical applications the compounds are advantageously formulated as creams or ointments or similar vehicle suitable for topical applications. Any such formulations may also contain other pharmaceutically-acceptable and non-toxic excipients such as stabilizers, anti-oxidants, binders, coloring agents or emulsifying or taste-modifying agents.

The compounds are advantageously administered by injection, or by intravenous infusion of suitable sterile solutions, or in the form of oral doses via the alimentary canal, or topically in the form of ointments, lotions, or in suitable transdermal patches. For the treatment of malignant diseases, the 1 9-nor vitamin D compounds of this invention are administered to subjects in dosages sufficient to inhibit the proliferation of malignant cells

and induce their differentiation into normal monocyte-macrophages. Suitable dosage amounts are from 1 to 500 µg of compound per day, as a single dose or individual dosages, such dosages being adjusted, depending on the disease to be treated, its severity and the response or condition of the subject as is well-understood in the art.

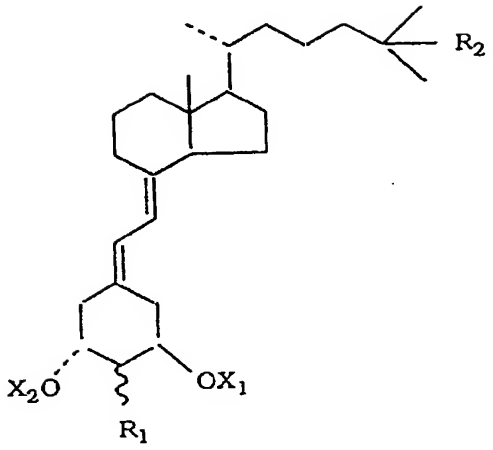
It can thus be seen that the compounds of this invention find utility in the treatment of senile or post menopausal osteoporosis as well as in low bone turnover osteoporosis. The compounds can be administered either during and subsequent to menopause or prior to the onset of menopause.

TABLE 1

Group	Amount pmoles/d/ 7 days	S/M Ratio ave. s.e.m.	Serum Ca ave. s.e.m. (mg %)
D-Deficient	0	$^{a}2.1 \pm 0.18$	$^{a}4.3 \pm 0.14$
1α,25-(OH)$_2$D$_3$	130	$^{b}8.0 \pm 0.80$	$^{b}5.6 \pm 0.20$
19-Nor-1α,2α, 25-(OH)$_3$ D$_3$	130	$^{c}5.3 \pm 0.26$	$^{1ca}4.12 \pm 0.3$
	325	5.0 ± 0.14	$^{cb}4.14 \pm 0.10$
D-Deficient	0	$^{a}3.0 \pm 0.8$	$^{a}4.3 \pm 0.23$
19-Nor-1α,2β, 25-(OH)$_3$D$_3$	65	$^{c}5.8 \pm 0.22$	$^{2ca}4.2 \pm .19$
	325	10.8 ± 0.39	$^{cb}4.2 \pm 0.28$
	650	9.9 ± 0.45	$^{cc}4.3 \pm 0.24$
1α,25-(OH)$_2$D$_3$	65	$^{b}8.3 \pm 0.32$	$^{b1}5.2 \pm 0.16$
	325	10.5 ± 1.2	$^{b2}6.8 \pm 0.24$
D-Deficient	0	$^{a}2.5 \pm 0.15$	$^{a}4.3 \pm 0.13$
1α,25-(OH)$_2$D$_3$	130	$^{b}7.1 \pm 0.49$	$^{b1}5.7 \pm 0.23$
	325	7.8 ± 0.60	$^{b2}7.1 \pm 0.30$
19-Nor-2α-hydroxy-propoxy-1α,25-(OH)$_2$D$_3$	130	$^{c}3.5 \pm 0.23$	$^{3ca}4.9 \pm 0.11$
	325	4.3 ± 0.33	$^{3b}5.8 \pm 0.20$
Statistical data Serosal/Mucosal Panel 1 All from a P < .001 Panel 2 All from a P < .001 Panel 3 All from a P < .001		Serum Ca Panel 1, b from a P < .001 $^{1c^a}$, c^b from a N.S. Panel 2, b^1, b^2, from a p<.001 $^{2c^a c^b c^c}$ N.S. Panel 3 b^1, b^2 from a p < .001 (3) c^a from a p < .0005 c^b from a, p< .001	

Claims

1. A vitamin D compound having the structure:



where X_1 and X_2 are each independently hydrogen or a hydroxy protecting group, R_1 which may be in either α or β position is a hydroxy group, protected hydroxy group or the group OR_3 where R_3 is an alkyl, hydroxyalkyl, fluoroalkyl, arylalkyl or aryl group, and R_2 is hydrogen or a hydroxy group.

2. 1α,2α,25-trihydroxy-19-nor-vitamin D_3.

3. 1α,2β,25-trihydroxy-19-nor-vitamin D_3.

4. 1α,25-dihydroxy-2α-(3'hydroxypropoxy)-19-nor-vitamin D_3.

5. 1α,25-dihydroxy-2β-(3'hydroxypropoxy)-19-nor-vitamin D_3.

6. 1α,25-dihydroxy-2α(benzyloxy)-19-nor-vitamin D_3.

7. A pharmaceutical composition comprising a 19-nor-vitamin D compound as claimed in any one of claims 1 to 6 together with a pharmaceutically acceptable excipient.

8. A pharmaceutical composition according to claim 7 wherein said 19-nor-vitamin D compound is 1α,2α,25-trihydroxy-19-nor-vitamin D_3, 1α,2β,25-trihydroxy-19-nor-vitamin D_3, 1α,25-dihydroxy-2α-(3'-hydroxypropoxy)-19-nor-vitamin D_3, 1α,25-dihydroxy-2β-(3'-hydroxypropoxy)-19-nor-vitamin D_3 or 1α,25-dihydroxy-2α-(benzyloxy)-19-nor-vitamin D_3.

9. A composition according to claim 7 or 8 wherein the 19-nor-compound is in solution in an ingestible and nontoxic liquid vehicle, said solution being encapsulated.

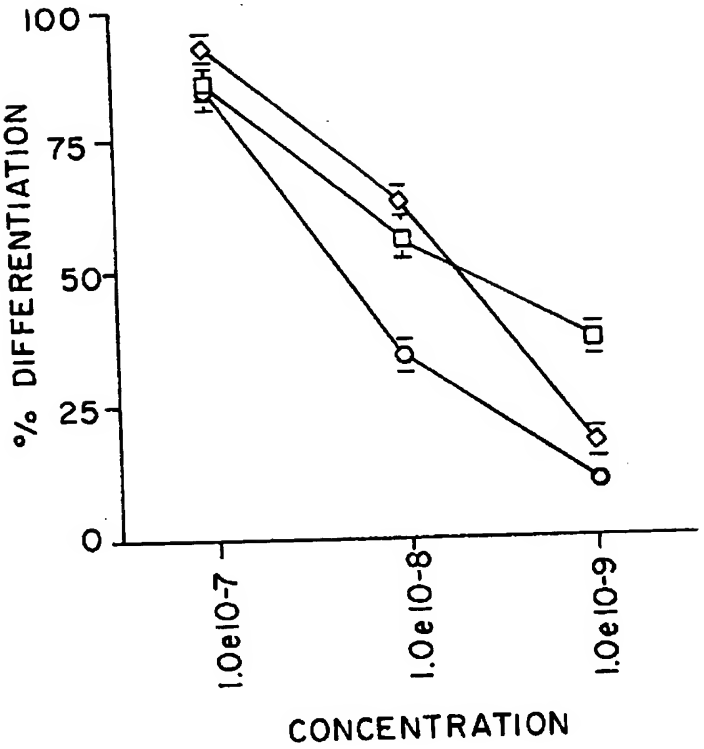
10. A composition according to claim 7 or 8 which is a slow release formulation.

11. A composition according to any one of claims 7 to 10 which contains 1 ug to 500 ug of the 19-nor-vitamin D_3 compound.

12. A vitamin D compound as defined in any one of claims 1 to 6 for use in treating osteoporosis.

13. The use according to claim 12 wherein the 19-nor-vitamin D compound is administered in an amount of from 1 ug to 500 ug per day.

HL-60 DIFFERENTIATION BY NBT ASSAY



—□— 1α, 25(OH)$_2$D$_3$

—◇— 19 NOR-1α25(OH)$_2$-2α(3 OH-PROPOXY)D$_3$

—○— 19 NOR-1α2α25(OH)$_3$D$_3$

FIG. 1A

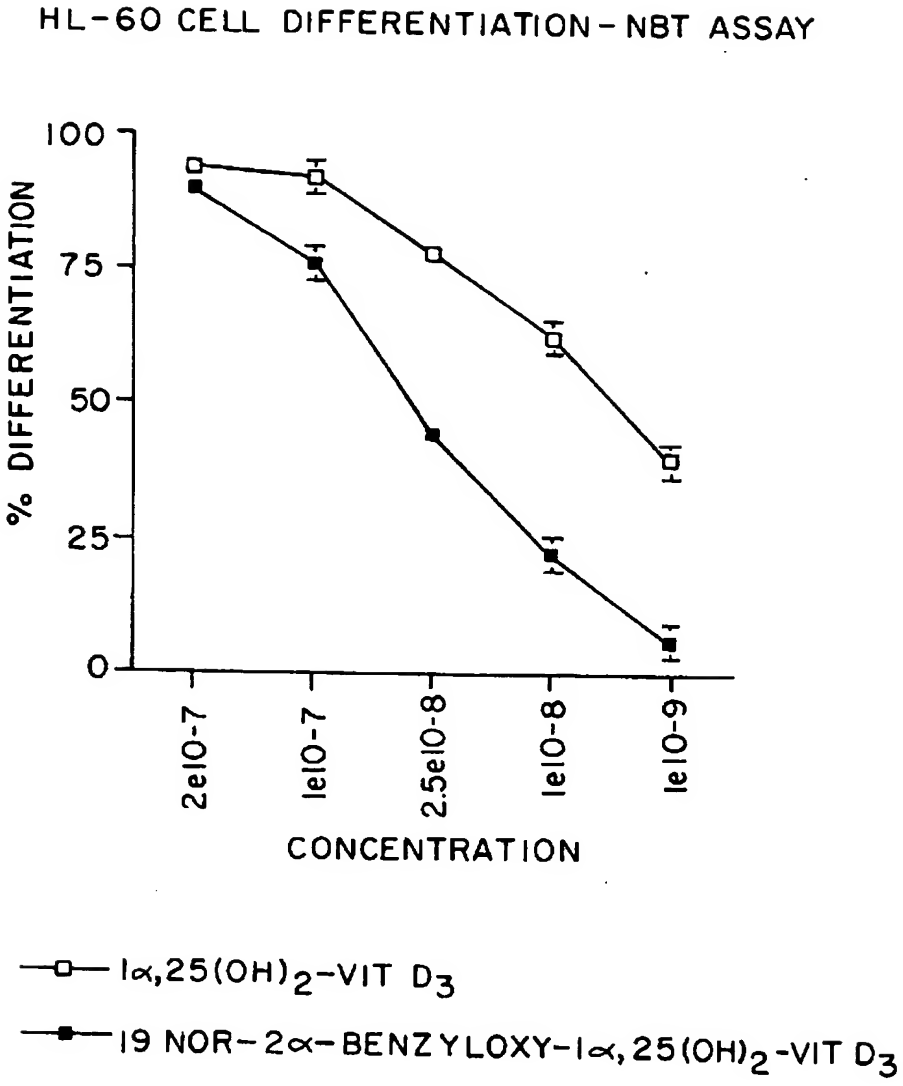

HL-60 CELL DIFFERENTIATION - NBT ASSAY
% DIFFERENTIATION
100
75
50
25
0
2e10-7
1e10-7
2.5e10-8
1e10-8
1e10-9
CONCENTRATION
1α,25(OH)2-VIT D3
19 NOR-2α-BENZYLOXY-1α,25(OH)2-VIT D3

FIG. 1B

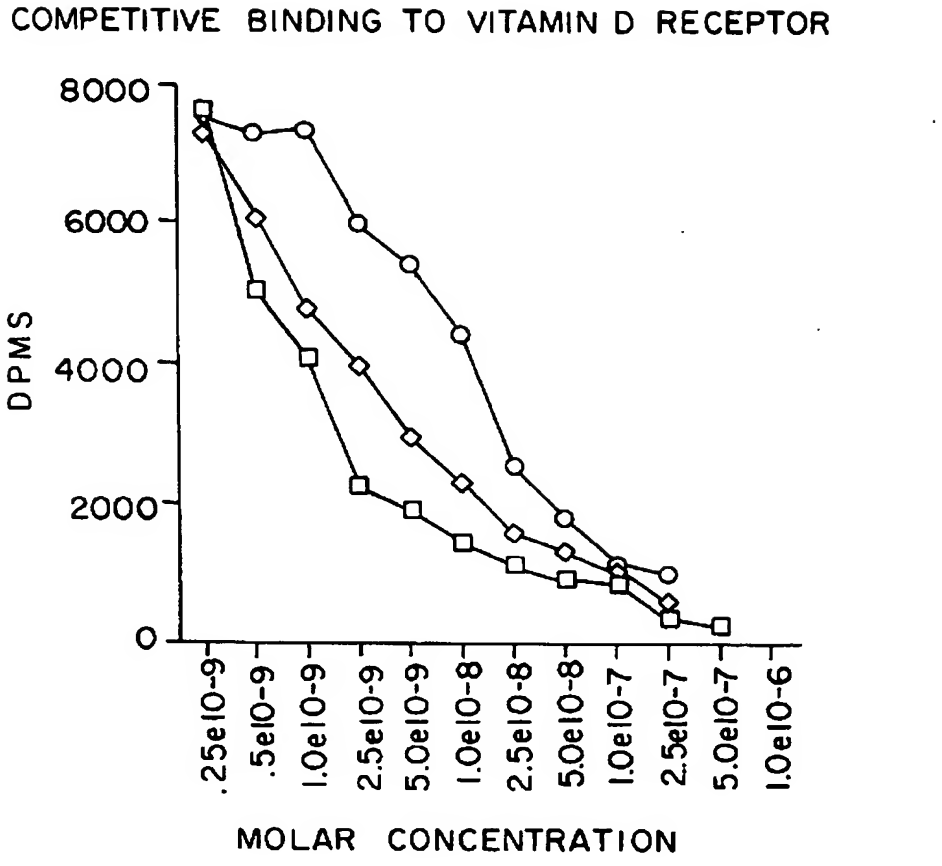

COMPETITIVE BINDING TO VITAMIN D RECEPTOR
8000
6000
4000
2000
0
DPMS
.25e10-9
.5e10-9
1.0e10-9
2.5e10-9
5.0e10-9
1.0e10-8
2.5e10-8
5.0e10-8
1.0e10-7
2.5e10-7
5.0e10-7
1.0e10-6
MOLAR CONCENTRATION
1α,25(OH)2VIT D3
19NOR-1α25(OH)2-2α(3OH-PROPOXY) VIT D3
19 NOR-1α,2α,25(OH)3 VIT D3

FIG. 2A

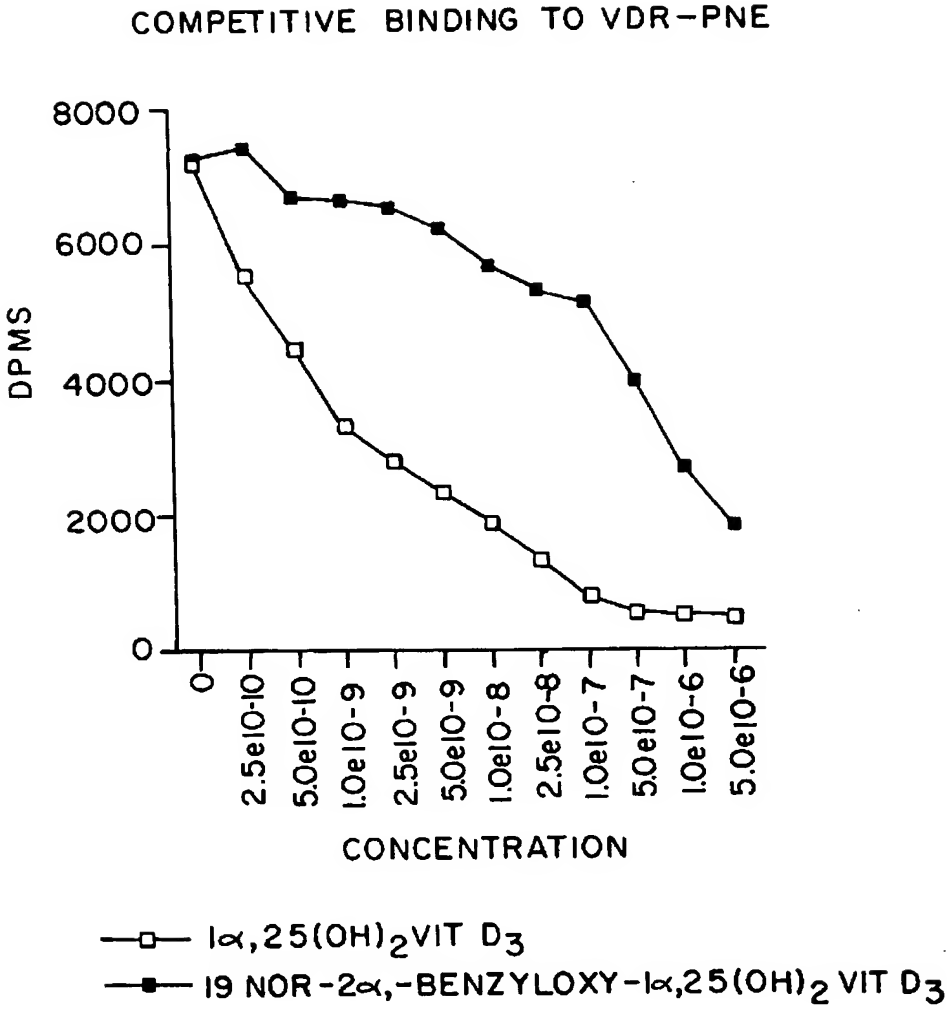

COMPETITIVE BINDING TO VDR-PNE
8000
6000
4000
2000
0
DPMS
0
2.5e10-10
5.0e10-10
1.0e10-9
2.5e10-9
5.0e10-9
1.0e10-8
2.5e10-8
1.0e10-7
5.0e10-7
1.0e10-6
5.0e10-6
CONCENTRATION
1α,25(OH)2VIT D3
19 NOR-2α,-BENZYLOXY-1α,25(OH)2 VIT D3

FIG. 2B

European Patent Office

EUROPEAN SEARCH REPORT

Application Number
EP 94 30 2382

DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (Int.Cl.5)
A	EP-A-0 387 077 (WISCONSIN ALUMNI RESEARCH FOUNDATION) * page 7, line 55 - page 8, line 24; examples 1e, 2 e; claims 1-4, 12-18 *	1,7	C07C401/00 A61K31/59
A	EP-A-0 516 410 (WISCONSIN ALUMNI RESEARCH FOUNDATION) * page 15, compound 20 *	1	
A D	EP-A-0 184 206 (CHUGAI SEIYAKU KK) * page 1, lines 3-10; examples 1-4, 6, 7, 9, 10, 13, 14; claims 1-5, 7 * & US-A-4 666 634	1,5,7,8, 12	
A	J. ORG. CHEM., vol.56, no.14, 1991 pages 4339 - 4341 G.H. POSNER ET AL * page 4339, summary *	1,2	
A	CHEMICAL ABSTRACTS, vol. 110, no. 10, 1989, Columbus, Ohio, US; abstract no. 82505y, * abstract * & JP-A-63 107 929 (CHUGAI PHARMACEUTICAL CO LTD)	1,5,7-9, 12	

TECHNICAL FIELDS SEARCHED (Int.Cl.5)
C07C
A61K

The present search report has been drawn up for all claims

Place of search	Date of completion of the search	Examiner
BERLIN	24 May 1994	Van Amsterdam, L

CATEGORY OF CITED DOCUMENTS

X : particularly relevant if taken alone
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L : document cited for other reasons

& : member of the same patent family, corresponding document

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